Safety, reactogenicity, and immunogenicity of a chimpanzee adenovirus vectored Ebola vaccine in adults in Africa: a randomised, observer-blind, placebo-controlled, phase 2 trial

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Summary

Background: The 2014 Zaire Ebola virus disease epidemic accelerated vaccine development for the virus. We aimed to assess the safety, reactogenicity, and immunogenicity of one dose of monovalent, recombinant, chimpanzee adenovirus type-3 vectored Zaire Ebola glycoprotein vaccine (ChAd3-EBO-Z) in adults.

Methods This phase 2, randomised, observer-blind, controlled trial was done in study centres in Cameroon, Mali, Nigeria, and Senegal. Healthy adults (≥18 years) were randomly assigned with a web-based system (1:1; minimisation procedure accounting for age, gender, centre) to receive ChAd3-EBO-Z (day 0), or saline placebo (day 0) and ChAd3-EBO-Z (month 6). The study was observer-blind until planned interim day 30 analysis, single-blind until month 6, and open-label after month 6 vaccination. Primary outcomes assessed in the total vaccinated cohort, which comprised all participants with at least one study dose administration documented, were serious adverse events (up to study end, month 12); and for a subcohort were solicited local or general adverse events (7 days post-vaccination), unsolicited adverse events (30 days post-vaccination), haematological or biochemical abnormalities, and clinical symptoms of thrombocytopenia (day 0–6). Secondary endpoints (subcohort; per-protocol cohort) evaluated anti-glycoprotein Ebola virus antibody titres (ELISA) pre-vaccination and 30 days post-vaccination. This study is registered with ClinicalTrials.gov, NCT02485301.

Findings Between July 22, 2015, and Dec 10, 2015, 3030 adults were randomly assigned; 3013 were included in the total vaccinated cohort (1509 [50·1%] in the ChAd3-EBO-Z group and 1504 [49·9%] in the placebo/ChAd3-EBO-Z group), 17 were excluded because no vaccine was administered. The most common solicited injection site symptom was pain (356 [48%] of 748 in the ChAd3-EBO-Z group *vs* 57 [8%] of 751 in the placebo/ChAd3-EBO-Z group); the most common solicited general adverse event was headache (345 [46%] in the ChAd3-EBO-Z group *vs* 136 [18%] in the placebo/ChAd3-EBO-Z group). Unsolicited adverse events were reported by 123 (16%) of 749 in the ChAd3-EBO-Z group and 119 (16%) of 751 in the placebo/ChAd3-EBO-Z group. Serious adverse events were reported for 11 (1%) of 1509 adults in the ChAd3-EBO-Z group, and 18 (1%) of 1504 in the placebo/ChAd3-EBO-Z group; none were considered vaccination-related. No clinically meaningful thrombocytopenia was reported. At day 30, anti-glycoprotein Ebola virus antibody geometric mean concentration was 900 (95% CI 824–983) in the ChAd3-EBO-Z group. There were no treatment-related deaths.

Interpretation ChAd3-EBO-Z was immunogenic and well tolerated in adults. Our findings provide a strong basis for future development steps, which should concentrate on multivalent approaches (including Sudan and Marburg strains). Additionally, prime-boost approaches should be a focus with a ChAd3-based vaccine for priming and boosted by a modified vaccinia Ankara-based vaccine.

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Introduction

Ebola virus disease is a serious illness with a case fatality rate between 25% and 90%.[1] The Ebola virus genus contains five distinct species of filoviruses. The Zaire Ebola virus was responsible for the largest 2014–16 Ebola virus disease epidemic: 28 616 Ebola virus disease cases were reported in Guinea, Liberia, and Sierra Leone, of which 11 310 were fatal.[2]

The risk of re-emergence of Ebola virus disease is illustrated by the 2017 and 2018–19 outbreaks in the Democratic Republic of the Congo.[3] A well tolerated and effective single-dose Ebola vaccine able to elicit rapid

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Research in context

Evidence before this study

We searched PubMed for clinical trials assessing Ebola vaccines from database inception up to Oct 14, 2019, with the title or abstract search terms "Ebola" AND "vaccine" AND "trial". No language restrictions or inclusion and exclusion criteria were applied. Four phase 1 clinical trials (assessing two DNA vaccines and one recombinant adenovirus serotype 5 vaccine) were done before 2014. All clinical trials evaluating the chimpanzee adenovirus type-3 vectored Zaire Ebola glycoprotein vaccine (ChAd3-EBO-Z) were prompted by the 2014–16 epidemic assessing various doses and further evaluating its safety and reactogenicity. Other viral vector-based vaccines evaluated after onset of the epidemic in 2014 included: rVSV-ZEBOV (a recombinant vesicular stomatitis virus-vectored vaccine expressing a Zaire Ebola virus glycoprotein); recombinant adenovirus type-5 (rAd5) vector or type-26 (rAd26) vector-based vaccines expressing the glycoprotein of the 2014 epidemic strain; modified vaccinia Ankara (MVA)-BN-Filo (a MVA vector vaccine encoding glycoproteins from Ebola virus, Sudan virus, and Marburg virus, as well as nucleoprotein from Tai Forest virus); MVA-EBO-Z (a recombinant, replication-deficient, attenuated vaccinia Ankara virus vector expressing the wild-type Ebola glycoprotein of the Zaire Mayinga strain); and GamEvac Combi (a vaccine consisting of rVSV and rAd5 expressing the envelope glycoprotein of the 2014 epidemic strain). Efficacy has only been evaluated and observed for the rVSV-ZEBOV in a phase 3 ring vaccination trial. Further efficacy assessments could not be done because of the end of the epidemic. So far, rVSV-ZEBOV is the only licensed vaccine.

Added value of this study

This is the largest trial with the ChAd3-EBO-Z investigational vaccine reported to date, to our knowledge. In this phase 2 study, we provided a comprehensive assessment of the vaccine, evaluating its safety, reactogenicity, and immunogenicity in terms of antibody response against Ebola virus glycoprotein, Ebola virus glycoprotein-specific T-cell response, and neutralising antibody response against the Ebola virus and ChAd3 vector. We found that the ChAd3-EBO-Z vaccine candidate had an acceptable safety and reactogenicity profile, in line with observations from phase 1 studies. In the phase 1 studies, transient decreases in thrombocyte counts had been observed; although not clinically meaningful, these observations were regarded as a potential safety signal. In the current phase 2 study, we did not observe meaningful trends towards decreased thrombocyte counts after vaccination, and no cases of clinically meaningful thrombocytopenia were observed within 7 days after vaccination. Antibody response against Ebola virus glycoprotein was induced after one dose, confirming observations from the phase 1 studies, and persisted up to 12 months post-vaccination. We also observed some polyfunctional Ebola virus-specific CD4+ and CD8+ T-cell responses.

Implications of all the available evidence

Several regimens of vaccine candidates against Ebola virus in clinical evaluation proved to have an acceptable safety profile and induced antibody responses. Safety data available to date support the clinically acceptable safety and reactogenicity profile of the investigational ChAd3-EBO-Z vaccine. We showed the immunogenicity of the vaccine with a single vaccination approach. The induced anti-glycoprotein Ebola virus immunoglobulin G amounts were substantial, although lower than those elicited by rVSV-ZEBOV 1 month post-vaccination (PREVAIL study) for which vaccine efficacy was observed. It was previously shown that ChAd3-EBO-Z vaccine is protective in a non-human primate model at the same dose as used in the current study. Although all available evidence concerning the protective potential of the vaccine against Ebola virus is indirect, data collected in the current phase 2 study will be useful in the development of similar vaccines against other Ebola strains.

protection would be a crucial tool to control future epidemics.[4] Current efforts target the Ebola virus glycoprotein.[5] Candidate DNA vaccines,[6–9] Ebola virus glycoprotein nanoparticle vaccine,[10] and candidate vaccines in which viral glycoprotein is expressed through various recombinant viral vectors have shown promising results in non-human primate models of Ebola virus disease[11–14] and in early phase clinical trials.[5,15–19] Recombinant vesicular stomatitis virus-based vaccine expressing Ebola virus surface glycoprotein (rVSV-ZEBOV) showed efficacy in a phase 3 ring vaccination trial in Guinea[20] and was granted conditional marketing authorisation by the European Medicines Agency in October, 2019.[21]

The investigational chimpanzee adenovirus type-3 (ChAd3)-vectored vaccine used in the present study, ChAd3-EBO-Z, is based on a replication-defective chimpanzee-derived adenovirus technology and contains a DNA fragment encoding Ebola virus glycoprotein. A single dose of 1×10^{11} particle units of ChAd3-EBO-Z provided 100% protection 4–5 weeks post-vaccination in a non-human primate model of Ebola infection.[14] Overall, the vaccine was well tolerated in phase 1 trials.[22–24] In some cases, thrombocyte counts transiently decreased, but no clinical signs or symptoms reported by participants suggested an increased tendency to bleed.[22–24]

This phase 2, randomised, placebo-controlled trial aimed to evaluate safety, reactogenicity, and immunogenicity following a single intramuscular dose of ChAd3-EBO-Z in adults in Africa and was evaluated in parallel in a phase 2 trial in children and adolescents in Mali and Senegal.[25]

Methods

Study design and participants

We did a phase 2, randomised, parallel-group, placebo-controlled, multicentre study at six centres in Africa: two in Cameroon, one in Mali, one in Nigeria, and two in Senegal

(appendix p 2). Eligible participants were healthy adults (≥18 years), as established by clinical examination at enrolment. Exclusion criteria included previous vaccination with investigational Ebola or Marburg vaccine, or a chimpanzee adenovirus-vectored vaccine; known previous Ebola virus or Ebola Sudan virus disease; travel to a country affected by the Ebola virus epidemic or direct contact with a person with Ebola virus disease within 21 days before the day 0 visit; and presence of any immunodeficiency state or any acute or chronic disease that was not well controlled, which could increase the risk for serious adverse events or could impair interpretation of the data. Fully detailed inclusion and exclusion criteria are provided in the appendix (p 1).

The study followed good clinical practice principles and the Declaration of Helsinki, with approval from national independent ethics committees (appendix p 1). Participants or their legally acceptable representatives provided written or thumb-printed informed consent. Participants younger than 21 years residing in Cameroon provided written or thumb-printed informed assent. Ten adult participants had an invalid informed consent form that could not be corrected and were excluded from all analyses; two participants (one in each group) had received the first dose. A protocol summary is available online.

Randomisation and masking

Participants were randomly assigned (1:1) at day 0 to the ChAd3-EBO-Z or placebo/ChAd3-EBO-Z group, using a central interactive response system. This interactive response system gave exclusive unmasked treatment information access to the unmasked centre personnel. The randomisation algorithm used a minimisation procedure accounting for age (18–40 years, 41–60 years, and >60 years), gender, and centre. Minimisation factors had equal weight in the minimisation algorithm. The determinism threshold was set at 80% (20% random allocation).

The study was observer-blind from study start until the planned interim day 30 analysis. Vaccine or placebo preparation and administration were done by authorised medical personnel who did not participate in any of the study clinical evaluation procedures. The study became single-blind (only the participant was unaware of treatment assignment) as of interim analysis until vaccination of the placebo/ChAd3-EBO-Z group at month 6, and open-label after month 6 vaccination. Some of the month 6 vaccinations occurred before the planned interim analysis, leading to unmasking. For these participants, it was ensured that the analysis of the day 0–30 data for interim analysis had been appropriately completed before unmasking.

Procedures

The ChAd3-EBO-Z group received ChAd3-EBO-Z at day 0; the placebo/ChAd3-EBO-Z group received placebo at day 0 and ChAd3-EBO-Z at month 6. Total study duration was approximately 12 months for each participant (figure 1). To obtain early data for ChAd3-EBO-Z in the context of accelerated vaccine development, a planned interim analysis was done when safety, reactogenicity, and immunogenicity data from all participants were available up to 30 days post-day 0 vaccination (data not shown). After this planned interim analysis, the study continued as planned.

ChAd3-EBO-Z consists of a recombinant replication-deficient adenovirus chimpanzee serotype 3 vector expressing wild-type Ebola virus glycoprotein from the Zaire Mayinga strain (dose 1×10^{11} particle units). The placebo control was phosphate-buffered saline. Study doses were administered intramuscularly in the deltoid of the non-dominant arm.

All participants were followed-up for serious adverse events throughout the study. Pregnancies and known pregnancy outcomes occurring during the study were recorded. A subcohort was followed-up during 7-day periods post-vaccination for solicited adverse events and 30-day periods post-vaccination for unsolicited adverse events. This subcohort included the first 750 participants enrolled in each group. Whole blood for analysis of laboratory safety parameters was drawn from all participants at screening, and from the subcohort for follow-up of adverse events at days 3, 6, 30, month 6, and month 12. Participants from the subcohort for follow-up of adverse events of the placebo/ChAd3-EBO-Z group had additional blood draws at month 6 +6 days and month 6 +30 days (figure 1, appendix p 3).

Because of transient non-clinically meaningful drops in thrombocyte counts observed in phase 1 trials, signs of clinically meaningful thrombocytopenia (ie, abnormal bleeding or increased tendency to bleed) starting within 7 days post-day 0 vaccination were assessed in the current study subcohort as an adverse event of special interest.

Anti-glycoprotein Ebola virus IgG antibody response was assessed by ELISA in the subcohort for adverse events. A blood sample was drawn at days 0 and 30, month 6, month 6 +30 days, and month 12 (figure 1). Serum was separated and frozen at –20°C. Antibody concentrations were determined using the Filovirus Animal Non-clinical Group ELISA at Q² Laboratories (San Juan Capistrano, CA, USA; appendix p 3). The technical cutoff was 36·11 ELISA units (EU)/mL, on the basis of the assay's lower limit of quantification. Antibody concentrations less than the cutoff were seronegative and given an arbitrary value of half the cutoff for geometric mean concentration calculation. Anti-glycoprotein antibody response was defined as a three-fold increase compared with baseline for participants with baseline concentrations of 36·11 EU/mL or more, or post-baseline seroconversion for baseline-seronegative participants. Seroconversion was defined as the appearance of antibodies (concentrations ≥36·11 EU/mL) in the serum of participants who were seronegative before vaccination.

See **Online** for appendix

For the **protocol summary** see https://www.gsk-studyregister.com/en/trial-details/?id=202091

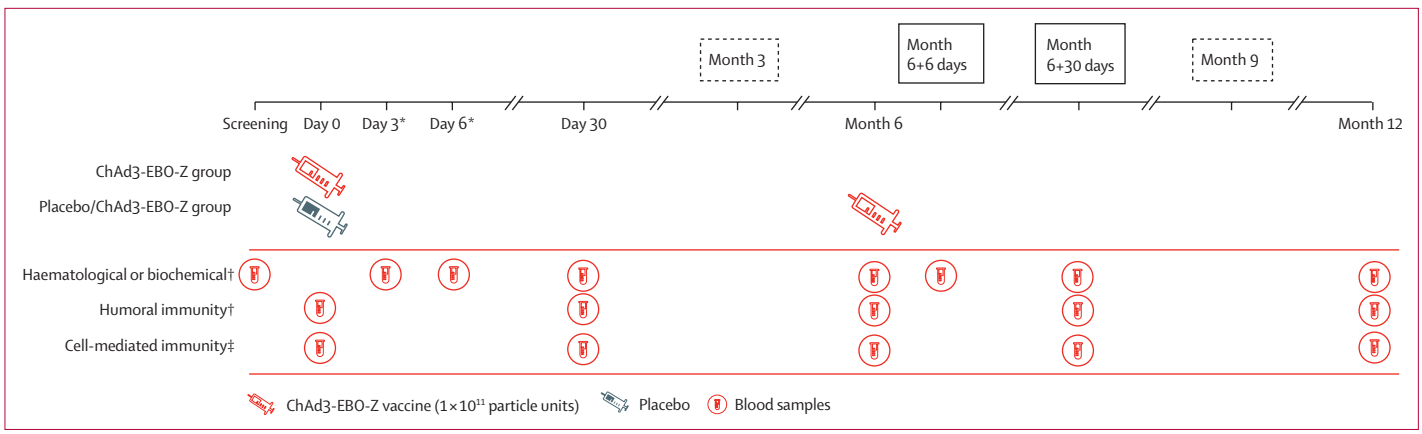

Figure 1: **Study design**
Dashed outlines indicate study contacts (home visit or phone call). Screening and day 0 visits were allowed to take place on the same day (allowed interval 0–30 days). Solid outlines (month 6 + 6 days and month 6 + 30 days) indicate visits only for participants in the subcohort for follow-up of adverse events and assessment of humoral immunity in the placebo/ChAd3-EBO-Z group.*These visits were only for participants in the subcohort for follow-up of adverse events and assessment of humoral immunity. †Blood samples only taken from participants in the subcohort for follow-up of adverse events and assessment of humoral immunity. ‡Blood samples were only taken from participants in the subcohort for cell-mediated immunity.

Additional immunological responses were evaluated in a subcohort, in which the first 100 enrolled participants per group were planned for Ebola virus glycoprotein-specific cell-mediated immunity and ChAd3 neutralising antibody (nAb) evaluation (figure 1). The Ebola virus glycoprotein-specific T-cell response was measured by intracellular cytokine staining (appendix p 4).

The Ebola virus neutralisation assay was done with infectious Ebola virus (Makona and Mayinga lineages) in biosafety level 4 facilities (Bernhard Nocht Institute for Tropical Medicine, Hamburg, Germany), using pre-vaccination and post-vaccination blood samples from 20 adult vaccinees (ten per group). Post-vaccination samples in the ChAd3-EBO-Z group were selected proportionally from the highest (three samples), lowest (three samples), and intermediate (four samples) concentrations of available anti-glycoprotein Ebola virus IgG antibodies. Serum from Ebola virus disease survivors and WHO reference serum for neutralisation assays were used as positive controls (appendix p 4).

The anti-ChAd3 neutralisation assay used a serotype-specific, replication-incompetent adenovirus, which has an inserted luciferase reporter gene, as described previously[26,27] and as detailed in the appendix (p 5). The threshold for positivity was set at a 90% nAb titre of 200.

Outcomes

The primary objective was to assess safety and reactogenicity of a single intramuscular dose of ChAd3-EBO-Z in healthy adults. The secondary objective was to evaluate humoral immunogenicity of a single intramuscular dose of ChAd3-EBO-Z, in terms of anti-glycoprotein Ebola virus IgG antibody responses as measured by ELISA.

The following primary endpoints were evaluated for a subcohort (approximately 750 participants per group): occurrence of solicited local or general adverse events within 7 days post-vaccination and unsolicited adverse events within 30 days post-vaccination; haematological and biochemical abnormalities at screening, days 3, 6, 30, months 6 and 12, and additionally at month 6 +6 days and month 6 +30 days for the placebo/ChAd3-EBO-Z group; and clinical symptoms of thrombocytopenia (day 0–6). Serious adverse events (up to month 12) were evaluated as the primary outcome for all participants. Secondary outcomes were anti-glycoprotein Ebola virus antibody concentrations (measured by ELISA) at day 0, day 30, month 6, and month 12; and percentage of seronegative or seropositive participants for anti-glycoprotein Ebola virus antibodies at day 0, day 30, month 6, and month 12. The secondary outcomes are further detailed in the appendix (p 2).

Persistence of antibodies, Ebola virus, and Ebola Sudan virus-glycoprotein specific T-cell responses, and nAb response against the ChAd3 vector, and further characterisation of immune response (Ebola virus nAb) after a single intramuscular dose of ChAd3-EBO-Z, were tertiary outcomes.

Statistical analysis

The study had no confirmatory objective and all analyses were descriptive. The target sample size of 1500 participants per group was calculated to fulfil the primary safety objective, according to consultations with regulatory authorities coordinated by WHO. The actual proportions associated with a 90% probability of observing a certain number of serious adverse events within a group of 1500 participants are provided in the appendix (p 5).

Safety analyses were done on the as treated cohort, and immunogenicity analyses in the per-protocol cohort.

Demographic characteristics were summarised by group using descriptive statistics for the total vaccinated cohort, which included all participants with at least one documented study dose administration. Safety and reactogenicity results (total vaccinated cohort) were

tabulated as the percentage of participants with a specific adverse event and its 95% CI (Clopper-Pearson method). We focused on the post-day 0 vaccination results since pooling of data after day 0 and month 6 ChAd3-EBO-Z vaccination did not alter conclusions regarding the vaccine's reactogenicity.

Immunogenicity was assessed for the per-protocol cohort comprising participants who met all eligibility criteria, had received at least one study dose according to protocol procedures and their random assignment, complied with protocol-defined procedures and intervals, and for whom data concerning immunogenicity endpoint measures were available.

For humoral immune response, seropositivity rates with exact 95% CIs were calculated by group. Geometric mean concentrations were tabulated, with 95% CIs as obtained by exponential transformation (base 10) of 95% CIs for the mean of the log-transformed concentrations. Mean geometric fold increases (MGI) were calculated as exponential transformation of the mean of the log transformed ratio of post-dose assay result to baseline assay result.

Vaccine responses to the antigen (with exact 95% CIs) were calculated. The same analyses were done by baseline anti-glycoprotein Ebola virus ELISA serological status. The relationship between day 30 anti-glycoprotein antibody ELISA titres (log values) and baseline anti-ChAd3 neutralisation titres (log values) was evaluated using a post-hoc linear regression, and expressed in terms of the correlation coefficient (r^2). For the cell-mediated immunity response, the frequency of specific CD4+ and CD8+ T cells was summarised for each study group using descriptive statistics.

All analyses were done using SAS, version 9.4. An unmasked independent data monitoring committee monitored safety and reactogenicity data. This study is registered with www.clinicaltrials.gov, NCT02485301 and was done in parallel with a phase 2 trial in children, NCT02548078.[25]

Role of the funding source

GlaxoSmithKline Biologicals SA was involved with study design, data collection, data analysis, data interpretation, and writing of the report. GlaxoSmithKline Biologicals SA paid for costs associated with the development and publishing of this manuscript. All authors had full access to all the data in the study, reviewed and commented on a draft version of the manuscript, and gave final approval before submission. All authors had final responsibility for the decision to submit for publication.

Results

Between July 22, 2015, and Dec 10, 2015, of 3770 screened adults, 3030 were randomly assigned to one of the study groups. The total vaccinated cohort included 3013 participants (1509 [50·1%] in the ChAd3-EBO-Z group and 1504 [49·9%] in the placebo/ChAd3-EBO-Z group; figure 2). Demographic and baseline characteristics were similar between the two groups (table 1). Most participants (3006 [99·8%]) were of African heritage, 1626 (54·0%) were male, and the median age was 29 years (range 18–85). 104 (3·4%) participants were withdrawn from the study (46 in the ChAd3-EBO-Z group and 58 in the placebo/ChAd3-EBO-Z group), mostly because of moving from the study area or loss to follow-up. One participant in the placebo/ChAd3-EBO-Z group erroneously received two doses of ChAd3-EBO-Z (at day 0 and month 6); the participant was excluded from immunogenicity analyses but did not report any serious adverse events after the second vaccination.

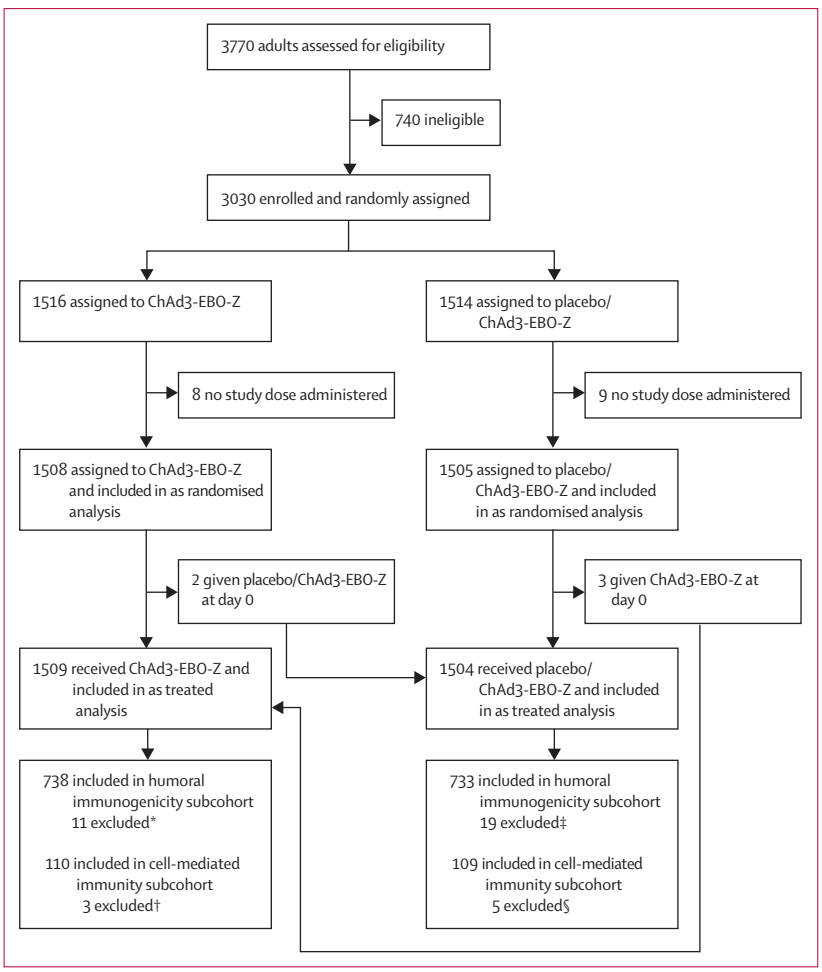

Figure 2: **Trial profile**

The ChAd3-EBO-Z group received ChAd3-EBO-Z vaccine at day 0, while the placebo/ChAd3-EBO-Z group received saline placebo at day 0 and ChAd3-EBO-Z at month 6. *11 excluded for: two laboratory results outside acceptable ranges; six non-compliance with visit schedule; one pre-existing prohibited medical condition; one received prohibited concomitant medication; one vaccine not administered per protocol. †Three excluded for: one laboratory results outside acceptable ranges; one received prohibited concomitant medication; one vaccine not administered per protocol. ‡19 excluded for (some for more than one reason): three laboratory results outside acceptable ranges; one excluded from the total vaccinated cohort; nine non-compliance with visit schedule; four pre-existing prohibited medical condition; one received prohibited concomitant medication; seven vaccine not administered per protocol. §Five excluded for (some for more than one reason): one excluded from the total vaccinated cohort; two non-compliance with visit schedule; two pre-existing prohibited medical condition; one vaccine not administered per protocol.

	ChAd3-EBO-Z (n=1509)*	Placebo/ChAd3-EBO-Z (n=1504)*
Subcohort		
Adverse events and humoral immunity	749 (49·6%)	751 (49·9%)
Cell-mediated immunity	112 (7·4%)	114 (7·6%)
Age, years		
Mean age	32·4 (18–85)	33·0 (18–84)
18–40	1136 (75·3%)	1133 (75·3%)
41–60	325 (21·5%)	327 (21·7%)
>60	48 (3·2%)	44 (2·9%)
Gender		
Women	697 (46·2%)	690 (45·9%)
Men	812 (53·8%)	814 (54·1%)
Health-care worker		
Yes	174 (11·5%)	176 (11·7%)
No	1335 (88·5%)	1328 (88·3%)
Race		
African heritage or African American	1505 (99·7%)	1501 (99·8%)
American Indian or Alaskan native	1 (0·1%)	0
Asian	3 (0·2%)	3 (0·2%)
Japanese heritage	1 (0·1%)	0
Southeast Asian heritage	2 (0·1%)	3 (0·2%)
Ethnicity		
American Hispanic or Latino	6 (0·4%)	11 (0·7%)
Not American Hispanic or Latino	1503 (99·6%)	1493 (99·3%)
Baseline childbearing potential of female participants		
Yes	547 (78·5%)	541 (78·4%)
No	150 (21·5%)	149 (21·6%)
Hysterectomy, bilateral ovariectomy, or current tubal ligation	21 (3·0%)	13 (1·9%)
Post-menopausal or pre-menarche	129 (18·5%)	136 (19·7%)
Physical characteristics		
Baseline height, cm	169·4 (142–205)	169·2 (145–200)
Baseline weight, kg	69·6 (40·0–122·0)	69·2 (38·9–142·5)
Country		
Cameroon	85 (5·6%)	85 (5·7%)
Mali	821 (54·4%)	823 (54·7%)
Nigeria	167 (11·1%)	163 (10·8%)
Senegal	436 (28·9%)	433 (28·8%)

Data are n (%) or mean (range). Baseline was the last non-missing assessment (scheduled or unscheduled) before the first vaccination at day 0. *Adults that were vaccinated at day 0 and month 6 with ChAd3-EBO-Z vaccine included.

Table 1: **Baseline characteristics of the total vaccinated cohort**

Solicited local symptoms within 7 days post-day 0 were reported by 358 (48%) of 748 participants in the ChAd3-EBO-Z group and 61 (8%) of 751 in the placebo/ChAd3-EBO-Z group. The most common solicited injection site symptom post-day 0 vaccination was pain, reported by 356 (48%) participants in the ChAd3-EBO-Z group and 57 (8%) participants in the placebo/ChAd3-EBO-Z group; most cases were grade 1 (figure 3). Grade 3 injection site pain was reported by three (<1%) of 748 ChAd3-EBO-Z recipients, all cases of which resolved within 4 days.

Solicited general symptoms within 7 days post-day 0 were reported by 450 (60%) of 748 participants in the ChAd3-EBO-Z group and 208 (28%) of 751 in the placebo/ChAd3-EBO-Z group. The most common solicited general symptom was headache, reported by 345 (46%) of 748 in the ChAd3-EBO-Z group and 136 (18%) of 751 in the placebo/ChAd3-EBO-Z group; most were grade 1 (figure 3). Grade 3 headache was reported by ten (1%) of 748 in the ChAd3-EBO-Z group and four (1%) of 751 in the placebo/ChAd3-EBO-Z group; nine cases in the ChAd3-EBO-Z group and three cases in the placebo/ChAd3-EBO-Z group were considered related to study vaccination by the investigator. Fever was reported by 106 (14%) of 748 in the ChAd3-EBO-Z group and 24 (3%) of 751 in the placebo/ChAd3-EBO-Z group, with a median duration of 1 day (SD 1·40, range 1–7) for the ChAd3-EBO-Z group and 1 day (1·32, 1–6) for the placebo/ChAd3-EBO-Z group. The highest incidence of fever occurred on day 1 post-vaccination (appendix p 6). Grade 3 fever was reported for three (<1%) of 748 ChAd3-EBO-Z recipients and was considered vaccination-related for two (<1%) participants.

At least one unsolicited adverse event within 30 days post-day 0 vaccination was reported by 123 (16%) of 749 participants in the ChAd3-EBO-Z group and 119 (16%) of 751 in the placebo/ChAd3-EBO-Z group (appendix p 7); most events were mild in severity. Eight severe unsolicited adverse events (two chills, two pain, one arthralgia, one myalgia, one back pain, and one polyuria) were reported by seven (1%) ChAd3-EBO-Z recipients; all events were considered vaccination-related. Two (<1%) participants in the placebo/ChAd3-EBO-Z group reported two unsolicited adverse events (ovarian cyst and tinnitus), of which one (tinnitus) was considered vaccination-related. The most frequently reported unsolicited adverse events in the ChAd3-EBO-Z group were malaria (n=31, 4%), rhinitis (n=16, 2%), increased alanine aminotransferase (n=15, 2%), anaemia (n=13, 2%), and dizziness (n=9, 1%); and in the placebo/ChAd3-EBO-Z group were malaria (n=31, 4%), rhinitis (n=25, 3%), increased alanine aminotransferase (n=16, 2%), and increased blood creatinine (n=12, 2%).

Changes in laboratory safety parameters are presented in the appendix pp 7–8 (ie, investigations and blood and lymphatic system disorders). Most events were mild in severity except two (<1%) ChAd3-EBO-Z recipients had moderate anaemia, one (<1%) participant in the placebo/ChAd3-EBO-Z group had a moderate increase in alanine aminotransferase, and one (<1%) participant in the placebo/ChAd3-EBO-Z group had a moderate increase in blood creatinine. Minor fluctuations from baseline

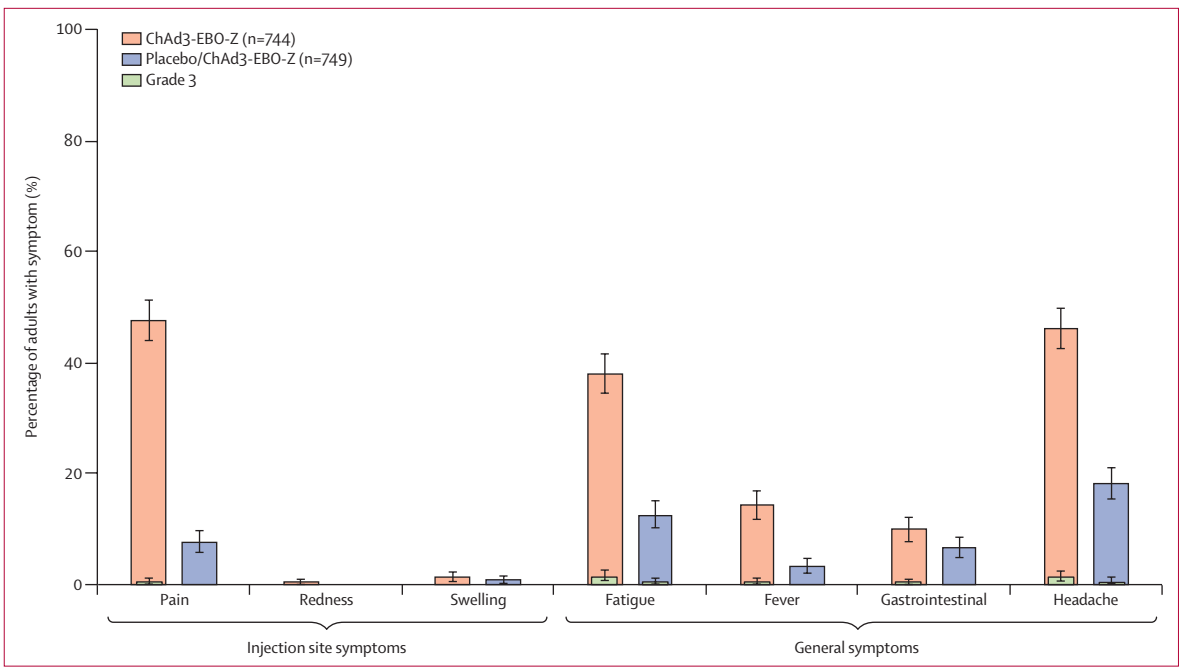

Figure 3: **Solicited injection site and general symptoms**
Data refer to the total vaccinated subcohort for adverse events and immunogenicity (day 0–6). Gastrointestinal symptoms included nausea, vomiting, diarrhoea, and abdominal pain. Error bars indicate 95% CIs.

thrombocyte concentrations were observed post-day 0 vaccination without notable differences between groups (appendix p 9).

No clinically meaningful thrombocytopenia within 7 days post-day 0 vaccination was reported for either group. Five mild cases of thrombocytopenia were reported as unsolicited adverse events within 30 days post-day 0 vaccination (appendix pp 7–8); two additional cases were reported post-month 6 ChAd3-EBO-Z vaccination. Pooled data after day 0 and month 6 ChAd3-EBO-Z vaccination did not differ from the post-day 0 reactogenicity and safety results (data not shown).

At least one serious adverse event was reported for 11 (1%) of 1509 participants in the ChAd3-EBO-Z group, and 18 (1%) of 1504 participants in the placebo/ChAd3-EBO-Z group of which three (<1%) occurred post-month 6 ChAd3-EBO-Z vaccination (appendix p 10); none were considered vaccination-related. Four participants died during the study. Two deaths occurred in the ChAd3-EBO-Z group: a 52-year-old man died after craniocerebral injury due to a motor vehicle accident 147 days post-vaccination; and a 29-year-old woman died 239 days post-vaccination due to rifampicin-induced liver injury. Two participants from the placebo/ChAd3-EBO-Z group who had received only the placebo, died during the study: one 26-year-old woman died 286 days after placebo administration due to postpartum haemorrhage, and one 19-year-old mother experienced the sudden death of a neonate with unknown cause, 413 days after placebo administration. None of these events were considered to be related to study vaccination.

46 (3·3%) of 1387 women reported pregnancies. One pregnant woman received placebo despite positive pregnancy urine test at screening; 14 women became pregnant after receiving ChAd3-EBO-Z, and 32 after receiving placebo (these women did not receive ChAd3-EBO-Z at month 6). The overall number of livebirths with no apparent congenital anomaly was 37 (80%) of 46 (12 [92%] of 13 in the ChAd3-EBO-Z group and 25 [76%] of 33 in the placebo/ChAd3-EBO-Z group); four elective and two spontaneous abortions were reported. One pregnancy resulted in early neonatal death, as detailed above under fatal serious adverse events.

No participants reported suspected Ebola virus disease during this study. At day 0 (before vaccination), 25% of participants had anti-glycoprotein Ebola virus IgG antibody concentrations of 36·11 EU/mL or greater (table 2). A reverse cumulative curve of the distribution of anti-glycoprotein Ebola virus IgG antibody concentrations at day 0 for participants with values of 36·11 EU/mL or greater at baseline (appendix p 11), showed that approximately 35% of participants had anti-glycoprotein concentrations greater than 200 EU/mL and 10% of participants had anti-glycoprotein concentrations greater than 1000 EU/mL. The proportion of participants with concentrations of 36·11 EU/mL or greater in the ChAd3-EBO-Z group was more than 97% at all post-vaccination timepoints, regardless of pre-vaccination concentrations (table 2). Anti-glycoprotein Ebola virus IgG antibodies persisted up to 12 months post-vaccination in the ChAd3-EBO-Z group, with 99% having

	Total			Baseline <36·11 EU/mL			Baseline ≥36·11 EU/mL		
	GMC (95% CI)	≥36·11 EU/mL n/N1 (%)	MGI (95% CI)	GMC (95% CI)	≥36·11 EU/mL n/N1 (%)	MGI (95% CI)	GMC (95% CI)	≥36·11 EU/mL n/N1 (%)	MGI (95% CI)
ChAd3-EBO-Z group									
Day 0	32 (29–35)	186/737 (25%)	NA	18 (18–18)	0/551	NA	174 (146–207)	186/186 (100%)	NA
Day 30	900 (824–983)	712/731 (97%)	28·1 (25·2–31·2)	795 (719–880)	530/545 (97%)	44·1 (39·8–48·7)	1300 (1089–1550)	181/185 (98%)	7·4 (6·1–9·1)
Month 6	459 (429–491)	716/721 (99%)	14·3 (13·0–15·9)	438 (406–472)	537/539 (100%)	24·3 (22·5–26·2)	528 (455–612)	178/181 (98%)	3·0 (2·5–3·7)
Month 6 +30 days	NA	NA	NA	NA	NA	NA	NA	NA	NA
Month 12	432 (402–465)	687/693 (99%)	13·6 (12·3–15·1)	405 (373–440)	520/523 (99%)	22·4 (20·7–24·4)	528 (454–615)	167/170 (98%)	2·9 (2·4–3·6)
Placebo/ChAd3-EBO-Z group									
Day 0	32 (29–35)	184/733 (25%)	NA	18 (18–18)	0/549	NA	176 (148–208)	184/184 (100%)	NA
Day 30	35 (32–38)	203/726 (28%)	1·1 (1·0–1·2)	21 (20–23)	46/543 (9%)	1·2 (1·1–1·3)	147 (120–180)	157/183 (86%)	0·9 (0·7–1·0)
Month 6	27 (25–28)	127/701 (18%)	0·8 (0·8–0·9)	20 (19–21)	23/522 (4%)	1·1 (1·1–1·1)	63 (51–77)	104/179 (58%)	0·4 (0·3–0·4)
Month 6 +30 days*	861 (796–931)	669/674 (99%)	26·6 (23·9–29·6)	798 (730–872)	501/504 (99%)	44·1 (40·4–48·2)	1079 (916–1270)	168/170 (99%)	6·1 (4·9–7·5)
Month 12*	566 (530–604)	670/672 (100%)	16·4 (14·8–18·2)	526 (488–568)	501/503 (100%)	27·2 (25·0–29·6)	700 (616–795)	169/169 (100%)	3·7 (3·1–4·5)

EU=ELISA units. GMC=geometric mean concentration. MGI=mean geometric fold increase. N1=total number of participants with results available at the relevant visit in the relevant analysis cohort per treatment group. NA=not applicable. Responders were defined as having a three-times increase in anti-glycoprotein antibody concentrations compared with baseline for participants with baseline concentration ≥36·11 EU/mL, or post-baseline seroconversion (appearance of antibody concentration ≥36·11 EU/mL) for baseline-seronegative participants. *The placebo/ChAd3-EBO-Z group received the ChAd3-EBO-Z vaccine at month 6; the results from month 6 + 30 days and month 12 should therefore be compared with the results of day 30 and month 6 in the ChAd3-EBO-Z group.

Table 2: **Seropositivity and anti-glycoprotein Ebola virus antibody GMC**

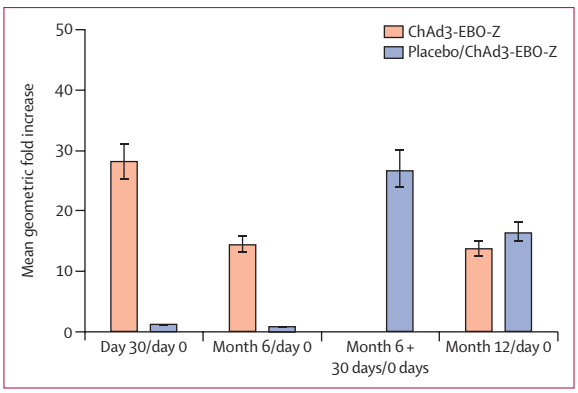

Figure 4: **Anti-glycoprotein ELISA mean geometric fold increase**
The ChAd3-EBO-Z group received the ChAd3-EBO-Z vaccine at day 0, and the placebo/ChAd3-EBO-Z group received the ChAd3-EBO-Z vaccine at month 6. The placebo/ChAd3-EBO-Z results from month 6 + 30 days and month 12 should therefore be compared with the results of day 30 and month 6 in the ChAd3-EBO-Z group. Error bars depict 95% CIs.

antibody concentrations of 36·11 EU/mL or greater (figure 4, table 2). Anti-glycoprotein Ebola virus IgG antibody geometric mean concentration in the ChAd3-EBO-Z group tended to be higher in participants with baseline titres of 36·11 EU/mL or greater (table 2). Anti-glycoprotein antibody responses at 30 days and 6 months post-ChAd3-EBO-Z vaccination in the placebo/ChAd3-EBO-Z group (thus, study month 6 +30 days, and study month 12) were similar to those observed at day 30 and month 6 in the ChAd3-EBO-Z group (table 3). At 30 days post-day 0 vaccination, anti-glycoprotein antibody response was observed for 92% of ChAd3-EBO-Z vaccinees, compared with 8% of participants who received placebo/ChAd3-EBO-Z (table 3). A graphical representation of anti-glycoprotein Ebola virus IgG immune responses is provided in the appendix (p 12).

Anti-glycoprotein ELISA MGI were highest at day 30 post-vaccination in the ChAd3-EBO-Z group, decreased until month 6, but remained stable by month 12, regardless of baseline serostatus. While MGI point estimates were much higher in adults with baseline anti-glycoprotein antibody concentrations of less than 36·11 EU/mL, the MGI value for adults with baseline concentrations of 36·11 EU/mL or greater was still 7·4 at day 30 (table 2). In the placebo/ChAd3-EBO-Z group, MGI values at 30 days and 6 months post-ChAd3-EBO-Z vaccination (month 6 +30 days and month 12) were similar to those observed at day 30 and month 6 in the ChAd3-EBO-Z group (figure 4).

Ebola virus glycoprotein-specific CD4+ and CD8+ T-cell responses were observed at day 30 post-vaccination (appendix pp 13–14). Ebola virus-specific CD4+ T-cell response was observed in 15 (36%) of 42 ChAd3-EBO-Z recipients at day 30 and 21 (21%) of 100 at month 6, compared with three (8%) of 39 at day 30 and 14 (14%) of 97 at month 6 in the placebo/ChAd3-EBO-Z group. Moderate increases of the proportion of Ebola virus-specific CD4+ T-cells were observed at 30 days post-ChAd3-EBO-Z vaccination, particularly for CD4+ T-cells co-expressing two (interleukin 2 [IL2] and tumour necrosis factor α [TNFα]) or three cytokines (IL-2, TNFα and interferon γ [IFNγ]). An Ebola virus-specific CD8+ T-cell response was observed in nine (21%) of 42 ChAd3-EBO-Z recipients at day 30, and 15 (15%) of 100 at month 6, compared with one (3%) of 39 at day 30 and 16 (16%) of 97 at month 6 in the placebo/ChAd3-EBO-Z group. When calculated for all participants from the cell-mediated immunity subcohort with results available at the relevant visit, the median percentage of Ebola virus

	Total		Baseline <36·11 EU/mL		Baseline ≥36·11 EU/mL	
	n/N1	Percentage (95% CI)	n/N1	Percentage (95% CI)	n/N1	Percentage (95% CI)
ChAd3-EBO-Z group						
Day 30	672/730	92% (89·9–93·1)	530/545	97% (95·5–98·5)	142/185	77% (70·0–82·6)
Month 6	633/720	88% (85·3–90·2)	537/539	100% (98·7–100)	96/181	53% (45·5–60·5)
Month 6 +30 days	NA	NA	NA	NA	NA	NA
Month 12	608/693	88% (85·1–90·1)	520/523	99% (98·3–99·9)	88/170	52% (44·0–59·5)
Placebo/ChAd3-EBO-Z group						
Day 30	57/726	8% (6·0–10·1)	46/543	9% (6·3–11·1)	11/183	6% (3·1–10·5)
Month 6	28/701	4% (2·7–5·7)	23/522	4% (2·8–6·5)	5/179	3% (0·9–6·4)
Month 6 +30 days*	621/674	92% (89·8–94·1)	501/504	99% (98·3–99·9)	120/170	71% (63·1–77·3)
Month 12*	603/672	90% (87·2–91·9)	501/503	100% (98·6–100)	102/169	60% (52·6–67·8)

EU=ELISA units. N1=total number of participants with results available at the relevant visit in the relevant analysis cohort per treatment group. NA=not applicable.
*The placebo/ChAd3-EBO-Z group received the ChAd3-EBO-Z vaccine at month 6; the results from month 6 + 30 days and month 12 should therefore be compared with the results of day 30 and month 6 in the ChAd3-EBO-Z group.

Table 3: **Anti-Ebola virus humoral immune response for anti-glycoprotein Ebola virus responders**

glycoprotein-specific CD8+ T cells expressing simultaneously IFNγ and TNFα (associated with protection in a preclinical non-human primate model) was 0·003% at day 0, 0·010% at day 30, 0·012% at month 6, and 0·008% at month 12 post-ChAd3-EBO-Z vaccination, and remained unchanged (0·0002% at each timepoint) for placebo. The percentage of Ebola virus glycoprotein-specific CD4+ and CD8+ T-cells expressing at least one cytokine, and co-expression profiles of Ebola Zaire antigen-specific CD4+ and CD8+ T cells are shown in the appendix (pp 13–14).

At day 30 post-ChAd3-EBO-Z vaccination, the MGI for Ebola virus glycoprotein-specific CD4+ T cells expressing at least one cytokine was 2·4 (95% CI 1·9–3·0); and for CD8+ T cells, 2·5 (1·8–3·3). At month 6 post-vaccination, MGI had decreased to 1·6 (95% CI 1·3–1·9) for CD4+ cells and 1·3 (1·0–1·8) for CD8+ cells. Groups are pooled together because of the low number of participants with available results (CD4+, 111 and 134 participants at 30 days and 6 months post-ChAd3-EBO-Z; CD8+, 110 and 131 participants at 30 days and 6 months post-ChAd3-EBO-Z). No Ebola Sudan virus glycoprotein-specific CD4+ or CD8+ T-cell responses were observed (data not shown).

None of the 20 selected participants (appendix p 4) had detectable neutralising antibodies against Ebola virus, with the lowest dilution titre that could be tested being 1/4 for the Makona lineage and 1/8 for the Mayinga lineage. No differences were observed between pre-vaccination and post-vaccination sera (data not shown). Positive controls were able to neutralise Ebola virus.

A ChAd3 nAb response above the threshold of positivity was observed in 32 (29%) of 110 participants at day 0, 63 (58%) of 108 at day 30, and 36 (35%) of 103 at month 6 in the ChAd3-EBO-Z group, and 32 (29%) of 109 participants at day 0, 28 (27%) of 103 at day 30, and 24 (25%) of 98 at month 6 in the placebo/ChAd3-EBO-Z group. In participants with pre-vaccination ChAd3 nAb titres greater than the threshold of positivity, ELISA antibody response to Ebola glycoprotein remained well above baseline until month 6. Based on a post-hoc linear regression analysis, ChAd3 nAb concentrations were found to explain only a small proportion (r^2=3%) of the ELISA anti-glycoprotein antibody titre variability. Post-day 30 in the ChAd3-EBO-Z group, anti-glycoprotein ELISA geometric mean concentrations were 670 (95% CI 475–844) in participants with baseline ChAd3 nAb greater than 200, and 850 (701–1030) in participants with baseline ChAd3 nAb less than 200 (appendix p 15). Anti-glycoprotein ELISA geometric mean concentrations by baseline anti-ChAd3 neutralisation status and baseline anti-glycoprotein ELISA seropositivity are shown in the appendix (p 16); however, only ten participants had baseline concentrations greater than cutoff for both anti-glycoprotein and anti-ChAd3. Groups were pooled together for increased accuracy, because of the low number of participants with available results.

Discussion

This phase 2 study provides a robust description of the safety, reactogenicity, and immunogenicity of the investigational ChAd3-EBO-Z vaccine, administered as a single intramuscular dose to healthy adults. The vaccine had an acceptable safety and reactogenicity profile, in line with observations from phase 1 studies.[22–24]

The most common solicited injection site symptom was pain (48%), with a similar incidence as the rVSV-EBOV phase 1 trial (57%)[28] and the rVSV-EBOV ring vaccination trial (47% at 0–30 min post-vaccination, 5·7% at 31 min–3 days post-vaccination).[20] The most common solicited general symptom was headache (46%), similar to the rVSV-EBOV phase 1 trial (47%)[28] but higher than in rVSV-EBOV ring vaccination trial (27% up to 30 min post-vaccination, 25% at 31 min–3 days post-vaccination).[20] Similar headache rates were observed for

licensed adjuvanted recombinant zoster vaccine (51% in adults 50–59 years), which had an even higher incidence of injection site pain (88%).[29]

While fever was reported by 14% of adults in the ChAd3-EBO-Z group, most cases were mild (≤38·5°C). All cases were short-lived and occurred immediately post-vaccination, which might help distinguish post-vaccination fever from fever due to Ebola virus disease in cases of mass vaccine deployment.

The potential effect of the vaccine on thrombocyte counts was assessed. Results confirmed the absence of decreasing trends in thrombocyte counts post-vaccination, and absence of clinically meaningful thrombocytopenia.

The vaccine induced an anti-glycoprotein Ebola virus antibody response at day 30, confirming findings from phase 1 trials, which used other ELISA assays.[22–24] We showed persistence of immune response up to 12 months post-vaccination, similar to results obtained in Liberia with the same vaccine and dose.[30] A persistent immune response is promising and desirable considering the length of time often required to control Ebola virus epidemics.

Pre-vaccination ChAd3 nAb concentrations explain only a small proportion of ELISA anti-glycoprotein antibody titre variability at day 30. Further investigation is needed to evaluate the effect of ChAd3 nAbs on the ELISA anti-glycoprotein response.

Ebola virus CD4+ and CD8+ T-cell responses observed after single vaccination were of low amplitude, although MGI at 30 days post-ChAd3-EBO-Z vaccination reached 2·4 for Ebola virus glycoprotein-specific CD4+ T cells expressing at least one cytokine, and 2·5 for CD8+ T cells. Similar low amounts of T-cell responses were previously observed after ChAd3-EBO-Z priming. Nevertheless, these responses were substantially boosted after vaccination with a modified vaccinia Ankara (MVA) vector encoding Ebola virus glycoprotein, even if T cells were not detectable at day 7 post-ChAd3-EBO-Z vaccination.[19,23,24] This observation suggests a true priming potential of the response obtained after single ChAd3-EBO-Z vaccination. Besides large-scale single vaccination approaches to contain epidemics in general populations, prime-boost approaches could be considered for specific populations requiring more durable protection (eg, health-care, front-line, and funeral workers).

Although individuals with known previous Ebola virus or Ebola Sudan virus disease were excluded from participation, approximately 25% had anti-glycoprotein Ebola virus IgG antibody concentrations of 36·11 EU/mL or greater before vaccination. This observation might result from potential assay limitations as the technical cutoff was determined using samples from a UK population. We recognise a risk of overestimation of anti-glycoprotein Ebola virus ELISA response, assuming that the cutoff is too sensitive for our study population. Nevertheless, the assay proved to clearly distinguish between pre-vaccinated and post-vaccinated individuals (both with and without high pre-vaccination ELISA reactivity). Even in ChAd3-EBO-Z vaccinees with baseline anti-glycoprotein antibody concentrations of 36·11 EU/mL and greater, MGI was 7·4 at 30 days post-vaccination.

In addition to potential assay limitations, we cannot completely rule out the hypothesis of some natural immunity against Ebola antigens or cross-antigens.[31] Zaire Ebola-specific antibodies have already been observed pre-vaccination[30,32,33] and might be ascribed to subclinical infections, and environmental or other factors. Seropositivity assessments outside of trials[34–37] and a serological survey testing oral swabs of asymptomatic household members of Ebola virus disease survivors confirmed existence of asymptomatic infection with Ebola virus, despite being uncommon.[38]

No nAb activity against Ebola virus was detected in our experimental conditions, including in samples with high ELISA anti-glycoprotein antibody activity. This finding contrasts with previous data showing nAb responses after single ChAd3-EBO-Z vaccination[23] and could be the result of less sensitive assay conditions in our study (appendix pp 4–5). Other potential factors include that our evaluation was done in an African population, not a UK population,[23] and the small number of specimens available for evaluation. Of note, even in the previous study, responses were of low amplitude but were boosted after vaccination with Ebola virus glycoprotein-encoding MVA.[23]

No correlate of protection against Ebola virus disease has yet been established for ChAd3-EBO-Z immune responses, and we could not show efficacy of ChAd3-EBO-Z in other study settings due to the rarefaction of Ebola cases in these settings.[30] Nevertheless, indirect evidence supports the protective potential of ChAd3-EBO-Z against Ebola virus disease, and hence a potential role in reactive vaccination to contain Zaire Ebola epidemics. ChAd3-EBO-Z was fully protective in non-human primate challenge experiments,[14] at the same dose as used in the current study in humans. Moreover, in another study in Liberia using an assay with the same technical specification, ChAd3-EBO-Z has been shown to induce anti-glycoprotein Ebola virus responses like those observed with the rVSV vaccine candidate,[30] for which efficacy was shown.[20] Moreover, we detected Ebola virus glycoprotein-specific CD4+ and CD8+ T-cell responses; CD8+ T-cell responses are considered to play a crucial role in conferring protection after adenovirus-based vaccination in non-human primate challenge experiments.[14,39] Although the observed T-cell responses after a single dose were low, this response is likely to be highly boostable, as supported by findings from previous studies.[23,24]

A plain language summary contextualising the results and their potential clinical relevance is provided in the appendix (p 17).

These data support the acceptable safety and reactogenicity profile of ChAd3-EBO-Z in adults. We observed anti-glycoprotein Ebola virus IgG antibody and

Ebola virus glycoprotein-specific CD4+ and CD8+ T-cell responses 30 days after single-dose vaccination, with antibody responses persisting up to 12 months post-vaccination. Long-term follow-up is needed for better characterisation of immunogenicity. The currently available information supports a role for single-dose vaccination in Ebola virus disease epidemic containment. The collected evidence, including a large and acceptable safety dataset in a wide study population (from the age of 1 year, when combined with the parallel paediatric study) can contribute to further improvements in ChAd3-EBO-Z and further vaccine development. Future development steps should concentrate on multivalent approaches, including Sudan and Marburg strains in the vaccine. Moreover, the focus should be on prime-boost approaches using ChAd3-based vaccine as priming and MVA-based vaccine as the booster, supported by encouraging results with this approach in previous studies.

Contributors

AN, PB, IDR, MD, MK, WRB, and FR designed the study. MDT, SOS, AT, BK, GAA, JA, SMb, BPN, CTN, KDM, KTTNK, GV, JJB, SO, KAK, KPA, and WRH collected the data. PB, EJ, IDR, MD, MK, WRB, FR, and SG did or supervised the analysis. AN, PG, PB, EJ, IDR, MD, MK, WRB, FR, and SG interpreted the data.

Zaire EBola Research Alliance group

Senate Amusu, Leo Ayuk, Catherine Bilong, Owusu Boahen, Makhtar Camara, Fadima Cheick Haidara, Daouda Coly, Siry Dièye, David Dosoo, Melanie Ekedi, Irma Eneida Almeida Dos Santos, Seyram Kaali, Afoke Kokogho, Myron Levine, Nick Opoku, Seth Owusu-Agyei, Simon Pitmang, Fatima Sall, Moussa Seydi, Marcelo Sztein, Mathurin Tejiokem, Awa Traore, Marie-Astrid Vernet, Abena Kunadu Yawson are part of the Zaire EBola Research Alliance group. All participated in data collection; however, they do not follow ICMJE conditions for authorship.

Declaration of interests

MD and IDR are employees of the GlaxoSmithKline (GSK) group of companies and report personal fees outside the submitted work. PB, EJ, MK, WRB, and FR are employees and hold restricted shares and stock options of the GSK group of companies. FR, MK, SG, and PB report grants from the European Commission and the EbolaVac grant, during the conduct of the study. IDR reports grants from International Consortium Emergency funds during the conduct of the study. KPA reports grants from Kintampo Health Research Centre during the conduct of the study. SOS, MDT, and GV report grants from GSK group of companies, during the conduct of the study. AN was an employee of Quintiles, a commercial entity that has received grants from the GSK group of companies and whom did part of the submitted work as a supplier to GSK. WRH is an employee of Q² Solutions which was responsible for the Ebola virus IgG ELISA. All other authors declare no competing interests. The views expressed in this work are the authors own and do not represent the views of the US Army or Department of Defense.

Data sharing

Within 6 months of this publication, anonymised individual participant data, annotated case report form, protocol, reporting and analysis plan, data set specifications, raw dataset, analysis-ready dataset, and clinical study report will be available for research proposals approved by an independent review committee. Proposals should be submitted online. A data access agreement will be required.

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